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Pattern of protein ingestion to maximise muscle protein synthesis after resistance exercise

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The maintenance of skeletal muscle mass is dependent upon the temporal and coordinated interaction between muscle/myofibrillar protein synthesis (MPS) and muscle protein breakdown (MPB). Resistance exercise (RE) alone elevates MPS and, to a lesser extent, MPB such that net muscle protein balance (NPB) remains negative. However, when RE is coupled with protein ingestion there is an accumulative effect on MPS resulting in a positive NPB (Phillips *et al.* 2005). Thus, repeated bouts of RE coupled with protein feeding is a viable strategy to maximise skeletal muscle hypertrophy and strength.

The impact of protein feeding on RE-induced increases in MPS has received much attention. One study has demonstrated that in young healthy males ∼20 g of high-quality protein is sufficient to maximise RE-induced rates of MPS over 4 h post-exercise (Moore *et al.* 2009). However, the interplay between the timing and quantity of protein consumed and subsequent anabolic responses throughout the course of a whole day is still poorly understood. In particular, there is a lack of data examining how the pattern of post-RE protein ingestion influences MPS later in the recovery phase (i.e. 4–12 h). A recent article published in *The Journal of Physiology* attempts to address this knowledge gap and in doing so provides valuable insights into how post-RE protein feeding strategies might be manipulated to optimise muscle anabolism. In an elegantly designed study, Areta *et al.* (2013) examined three groups of eight healthy, trained males. Participants performed a bout of bilateral leg extension RE followed by the consumption of 80 g of whey protein over 12 h of recovery ingested as either 8 × 10 g every 1.5 h, 4 × 20 g every 3 h or 2 × 40 g every 6 h. A stable isotope infusion was coupled with frequent skeletal muscle biopsy sampling to determine rates of MPS for 12 h post-RE. The data demonstrate that although all feeding strategies elevated MPS during the 12 h recovery period, consuming 20 g of whey protein every 3 h was the superior strategy for stimulating MPS rates. The authors concluded that these findings have the potential to maximise outcomes of resistance training designed to elicit a maximal hypertrophic response.

The data of Areta *et al.* show that manipulating the pattern of protein ingestion following RE can have a significant impact on the subsequent muscle anabolic response. The divergent feeding strategies of Areta *et al.* were used to mimic possible patterns of protein intake commonly observed in resistance-trained athletes. That is, 8 × 10 g every 1.5 h represents a 'grazing' approach, whereas 2 × 40 g every 6 h relates to the 'three square meals per day' approach. Yet, both of these strategies were inferior for stimulating MPS over 12 h of post-RE recovery compared with 4 × 20 g ingested every 3 h. However, it is important to note that this response was characterised when protein was ingested alone, and as the authors acknowledge, this finding cannot be evaluated in the context of a mixed meal. Indeed, it is commonplace to consume protein in the form of a mixed-macronutrient meal. Therefore, it is reasonable to postulate that macronutrient co-ingestion could alter intestinal transit, thus influencing amino acid absorption kinetics (Deutz *et al.* 1995) and perhaps MPS. Moreover, this study used high-quality whey protein and it remains to be seen if a similar pattern of MPS post-RE would be observed using the same feeding strategies with a slow-release protein such as casein. Such information may be valuable to individuals who choose not to (or are unable to) ingest high-quality protein in supplemental form following exercise, but instead consume whole-food protein sources.

Areta *et al.* should be highly commended for underlining the importance of not only the quantity, but particularly the pattern of post-RE protein ingestion to maximise the rate of MPS over 12 h. However, as a note of caution, their findings are limited to a healthy young male population. In this regard, recent evidence demonstrates that the elderly require more protein (40 to > 20 g) to elicit optimal increases in RE-induced rates of MPS than the young (Yang *et al.* 2012). It is therefore reasonable to consider whether the temporal influence of post-RE protein feeding on elderly muscle could be different compared to that of young. In this regard, the next logical step is to apply the model of Areta *et al.* in elderly and other populations, in whom maintenance of muscle mass is a critical determinant of longevity and quality of life. Yet, it should be acknowledged that Areta *et al.* afford data pertaining to only 12 h of recovery from RE. Hence, whether the acute responses of MPS to RE and protein feeding translate into a long-term functional response remains unknown.

The findings of Areta *et al.* will no doubt also grasp the attention of coaches and athletes alike. As such, some may cite the use of a bilateral exercise stimulus and absence of participants with large amounts of lean mass (>75 kg) as issues that preclude full applicability in a 'real-world' setting. To date, it is unclear whether exercising a greater volume of muscle mass is limiting for MPS in response to a given protein dose. Therefore, individuals with greater muscle mass or those engaged in whole-body RE training sessions may require ingestion of a greater protein dose to stimulate MPS maximally. With regard to the notion of applicability to the 'real-world' setting, it also may be significant that the participants entered the experimental trial in the fasted state. As a result the authors are unable to identify whether a pre-exercise meal would influence the MPS response to RE and various feeding strategies. This point becomes more relevant when considering the impact of insulin on MPB with regard to the true *growth* response and therefore the long-term applicability of the findings. Future studies assessing MPS and MPB in both the clinical and the athletic setting following RE and feeding are now required.

The study by Areta *et al.* also reveals novel nutrient–exercise interactions in cellular signalling. Phosphorylated mTOR^{Ser2448} was ∼2- to ∼6-fold above resting values throughout the 12 h recovery period independent of protein feeding strategy. Phosphorylation of p70S6K^{Thr389} was also increased above baseline, again in all feeding

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strategies. However, there was discordance between the degree of p70S6K^{Thr389} phosphorylation and the MPS response. In fact, the magnitude of phosphorylated p70S6K^{Thr389} displayed a 2×40 g to $> 4 \times 20$ g to $> 8 \times 10$ g pattern at 1 and 7 h post-RE. This finding is surprising given that phosphorylated p70S6K^{Thr389} is a key player in protein synthesis yet it was the 4×20 g strategy that induced the most favourable influence on MPS but median impact on phosphorylated p70S6K^{Thr389}. However, it is important to recognise that the timing of the biopsies at 1 and 7 h coincided with a greater volume of protein consumed prior to those biopsies for the 2×40 g condition, which may explain the discordance between p70S6K^{Thr389} signalling and MPS.

The common method employed to assay protein phosphorylation, a proxy of activity, in an exercise science setting, and in the present investigation, is Western blotting (WB). In contrast to the quantitative and reproducible techniques used to measure MPS, WB is a semi-quantitative method. Additionally, phosphorylated p70S6K^{Thr389} is recognised as a key controller of ribosomal biogenesis. So although the phosphorylation of p70S6K^{Thr389} post-RE does not correspond to the greatest acute MPS response it may in fact be leading to greater levels of ribosomal transcription. Interestingly, phosphorylation of p70S6K following RE often occurs in the nucleus, where ribosomal biogenesis commences. A caveat of the field is that no study has employed cellular fractionation techniques to reveal whether different RE and feedings strategies alter the ratio of nuclear to cytoplasmic phosphorylated p70S6K in human skeletal muscle. Hence, the lack of concordance between the MPS and signalling response in this and numerous other works emphasises the need for the development of new measures regarding readouts of ribosomal biogenesis in addition to fully quantitative methods to ascertain signalling activity following RE and nutrition.

To conclude, the study by Areta *et al.* contributes novel data to the body of literature highlighting the importance of the timing and quantity of protein consumed post-RE for muscle anabolism. By mimicking the habitual feeding strategies of many athletes engaged in resistance training, the authors move closer to bridging the gap between science and the applied setting. Future work that identifies the impact of different macronutrients consumed in combination, i.e. fat, carbohydrate, protein and fibre, on MPS in both elderly and young is warranted. Furthermore, there is growing interest in whether having greater amounts of muscle mass, or indeed exercising muscle mass involved in trainin impact RE-induced rates of MPS. Thus, future studies that examine the MPS response in individuals with large muscle mass, performing real-world RE, may provide informative data for clinical and athletic practice.

References

Areta JL, Burke LM, Ross ML, Camera DM, West DW, Broad EM, Jeacocke NA, Moore DR, Stellingwerff T, Phillips SM, Hawley J & Coffey VG (2013). Timing and distribution of protein ingestion during prolonged recovery from resistance exercise alters myofibrillar protein synthesis. *J Physiol* **591**, 2319–2331.

Deutz NEP, Ten Have GAM, Soeters PB & Moughan PJ (1995). Increased intestinal amino-acid retention from the addition of carbohydrates to a meal. *Clin Nutr* **14**, 354–364.

Moore DR, Robinson MJ, Fry JL, Tang JE, Glover EI, Wilkinson SB, Prior T, Tarnopolsky MA & Phillips SM (2009). Ingested protein dose response of muscle and albumin protein synthesis after resistance exercise in young men. *Am J Clin Nutr* **89**, 161–168.

Phillips SM, Hartman JW & Wilkinson SB (2005). Dietary protein to support muscle anabolism with resistance exercise in young men. *J Am Coll Nutr* **24**, 134S–139S.

Yang Y, Breen L, Burd NA, Hector AJ, Churchward-Venne TA, Josse AR, Tarnopolsky MA & Phillips SM (2012). Resistance exercise enhances myofibrillar protein synthesis with graded intakes of whey protein in older men. *Br J Nutr* **108**, 1780–1788.

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